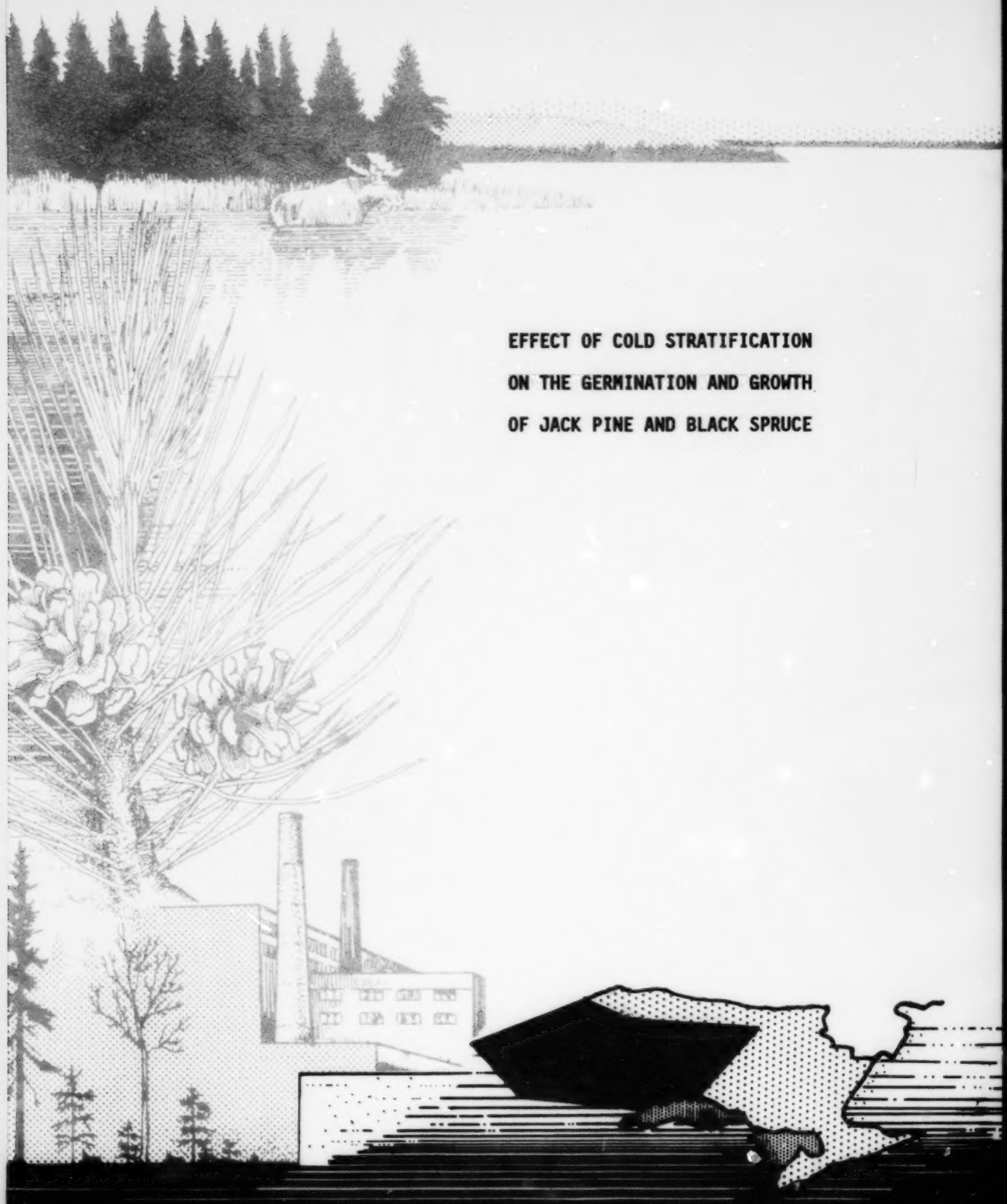
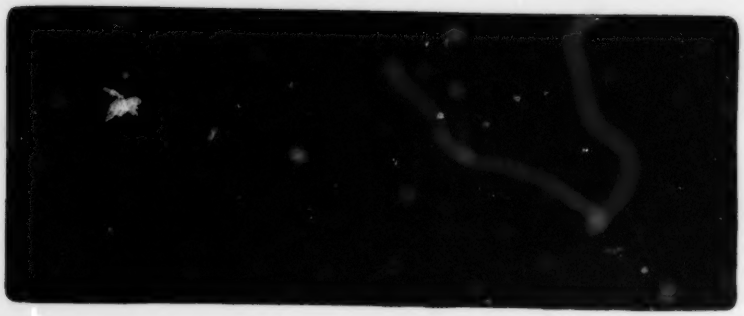


EFFECT OF COLD STRATIFICATION ON THE GERMINATION AND GROWTH OF JACK PINE AND BLACK SPRUCE

Ministry of Natural Resources
Ontario

NORTHWESTERN ONTARIO FOREST TECHNOLOGY DEVELOPMENT UNIT



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EFFECT OF COLD STRATIFICATION ON THE GERMINATION AND GROWTH OF JACK PINE AND BLACK SPRUCE

by

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INTRODUCTION

In an effort to improve the cost effectiveness of forest operations, seeding is gaining favour as a method of regenerating forest lands. Direct seeding allows regeneration of large areas, requires little labour and capital inputs relative to planting and can be done in spring or fall. Low survival rates, especially for black spruce (*Picea mariana* (Mill.) B.S.P.), and the enormous quantities of seed used in operational seeding have resulted in increased efforts to find practical methods of improving seed use efficiency and post seeding survival. Reducing seed use to two or three seeds per established seedling will help make the use of genetically improved seed feasible.

Seed conditioning treatments are used to improve germination rate and increase the survival of germinants (Boyer et al. 1985). Stratification, or moist, cold storage (Allen 1967; Bonner 1984), is used to overcome the internal dormancy exhibited by seeds of some species. For non-dormant species, stratification is used to increase the speed and uniformity of germination and thus increase field survival (Edwards 1986). For example, Verheggen (1982) found that black spruce seed stratified for three weeks at 4°C germinated four days earlier than non-stratified seed. Stratification may also reduce differences in germination rate caused by handling practices and offset the temperature and light requirements of germination (Farmer et al. 1984, Wang 1987).

Stratification results in faster, more uniform germination but requires time, and can magnify damage to seed caused by improper handling and storage if done improperly (Bonner et al. 1974, Edwards 1987). Seeds must be sown soon after removal from stratification or subjected to controlled drying; if they dry out too much they may die or go into deeper dormancy (Bonner et al. 1974). The response to drying is species and possibly seed lot specific.

Stratification is achieved by storing moistened seeds for given time periods at low temperatures. The requirements for successful stratification are: moisture to rehydrate tissues; cool temperatures to allow biochemical and morphological changes but delay sprouting, microorganism growth and overheating; aeration to minimize carbon dioxide accumulation and overheating; and correct duration (Edwards 1986, Wang 1987).

In Ontario, no standards exist for seed stratification periods (Wang 1989, pers. comm.). For black spruce, 2 - 4 weeks have been considered adequate (Farmer et al. 1984, Wang 1989, pers. comm.). Trials at Wabigoon nursery (Dryden, Ontario) show that 7 - 14 days of stratification is most effective in increasing germination rate but does not affect the number of germinants. Results in seedbeds were not as clearcut as in the germinator (Myland et al. 1988, Myland and Horvath 1988). Stratification periods longer than three months are detrimental to black spruce seed (McGillvray 1955).

For jack pine (*Pinus banksiana* Lamb.), 2 - 3 weeks is thought to be the most effective stratification time (Wang 1989, pers. comm.). Trials at Wabigoon nursery indicated seven day stratification treatments produced optimum results in the germinator but field trials were inconclusive (Myland et al. 1988, Myland and Horvath 1988).

Stratification has traditionally been done in bareroot nurseries prior to sowing (Bonner et al. 1974). Few trials using seed stratification in conjunction with operational aerial or ground seeding are documented in the literature. Treatments that increase germination speed may decrease the time that germinating seeds are subject to adverse conditions in the field and thus increase seeding success (Boyer et al. 1985; Bourgeois and Malek 1991). The increased use of direct seeding and shelter cones for regeneration warrants

testing stratification as a means of improving seeding success. Potential decreased seed requirements and increased time for growth and development would be advantageous to operational seeding programs.

The objectives of this study were:

1. To evaluate the effect of seed stratification on the germination and growth of jack pine and black spruce in the field.
2. To determine if seeding can be extended later into the summer using stratified seed.
3. To determine the effect of shelter versus bare seeding on stratified seed.

This report summarizes second and third year results of stratified seeding trials established north of Thunder Bay in 1989 and 1990.

METHODS

The trial included two species, black spruce and jack pine; two sowing methods, shelter seed and bare seed; four stratification treatments in 1989, 0 (control), 7, 15, and 30 days; three stratification treatments in 1990, 0, 7 and 15 days; four sowing dates in 1989, May, June, July, and August; and three sowing dates in 1990, May, July and August. Photodegradable cerkon cones were used as shelters.

Seeds

The seeds used in this experiment came from the Angus seed extraction plant. The black spruce seed (seed source 3425003) used in 1989 was collected in 1981 and tested at 97% viable in germination tests. The jack pine seed (seed source 3425001) was collected in 1983 and was also 97% viable.

The black spruce seed used in 1990 was collected in 1986 and tested at 96% viable. The jack pine seed was collected in 1988 and tested at 88% viable.

Stratification

Stratification treatments were accomplished by placing moistened seed on moist Kimpak paper in plastic bags in a seed cooler at 1-4°C for the specified time period of seven, fifteen or thirty days, such that all seeds for a seeding time were removed at the same time one day prior to sowing. During the stratification period, bags were opened once a week to aerate the seeds. The day before sowing, seeds were surface dried, bagged and kept refrigerated until sowing. This process was repeated for each sowing time. Control seeds were kept dry and refrigerated.

1989 Field Trial

In 1989, the field trial was located on Canadian Pacific Forest Products F.M.A. license near Camp 45 northeast of Thunder Bay (Figure 1). The site, which formerly supported an upland black spruce-jack pine mixture, was cut in 1987 and site prepared with a disc trencher in 1988. The site was classified as a Northwestern Ontario Forest Ecosystem Classification (NWO FEC) soil type S3 (fresh coarse-loamy soil) (Sims et al. 1989) with gently rolling topography.

Seeds were sown at four week intervals on: May 9 (jack pine) and May 11 (black spruce), June 6, July 4 (except 7 day seed on July 11), and August 1. Twenty five spots were sown for each species-treatment combination in each of seven blocks. The layout of the field trial is shown in Figure 2.

Seed spots were scuffed with boots to remove excess duff and debris. Shelter cones or spot markers were placed on the microsite and seeds were counted and sown into cones or near markers. Five seeds were sown under each shelter cone, ten were sown on bare seed spots.

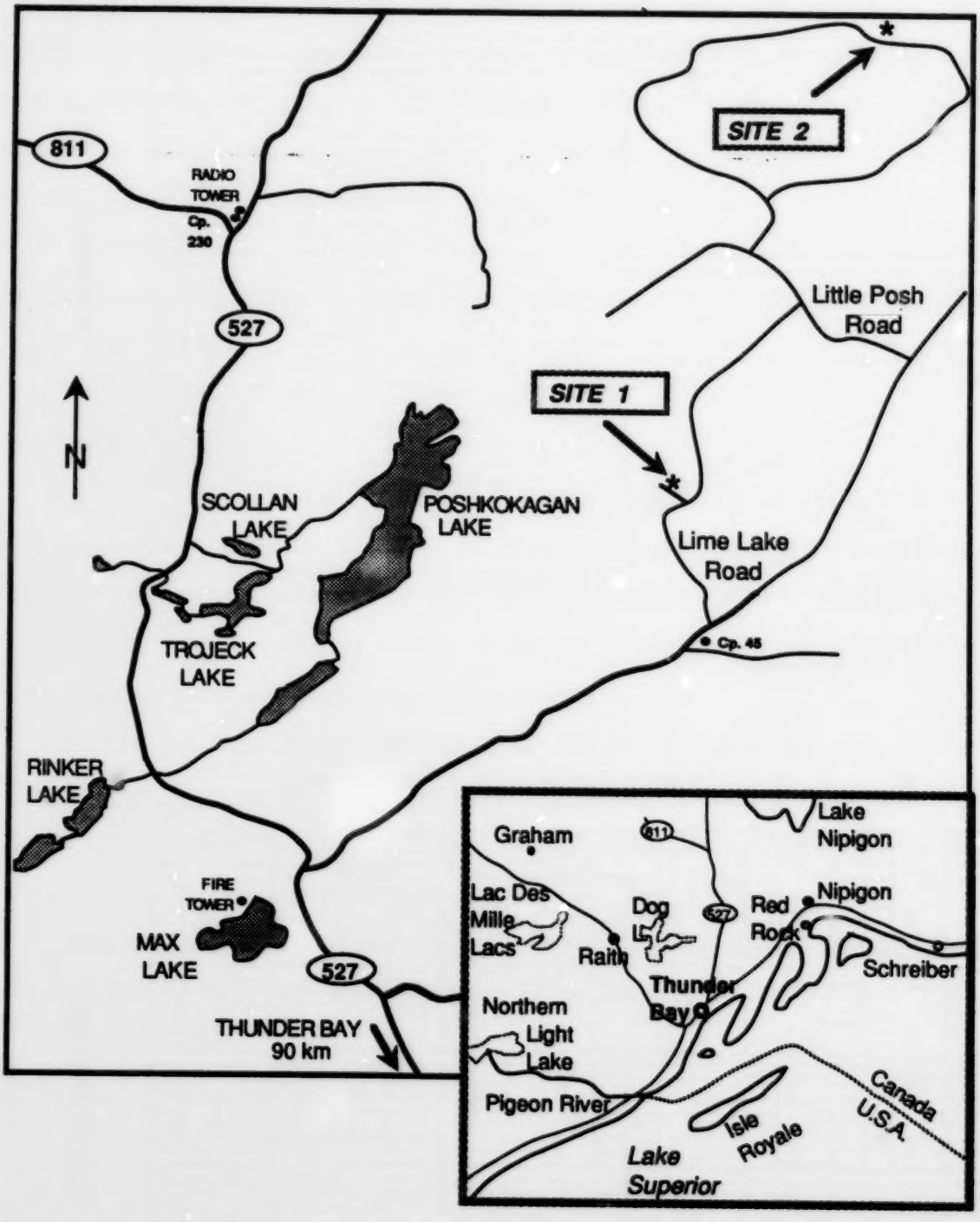

Figure 1. General location of field trial sites.

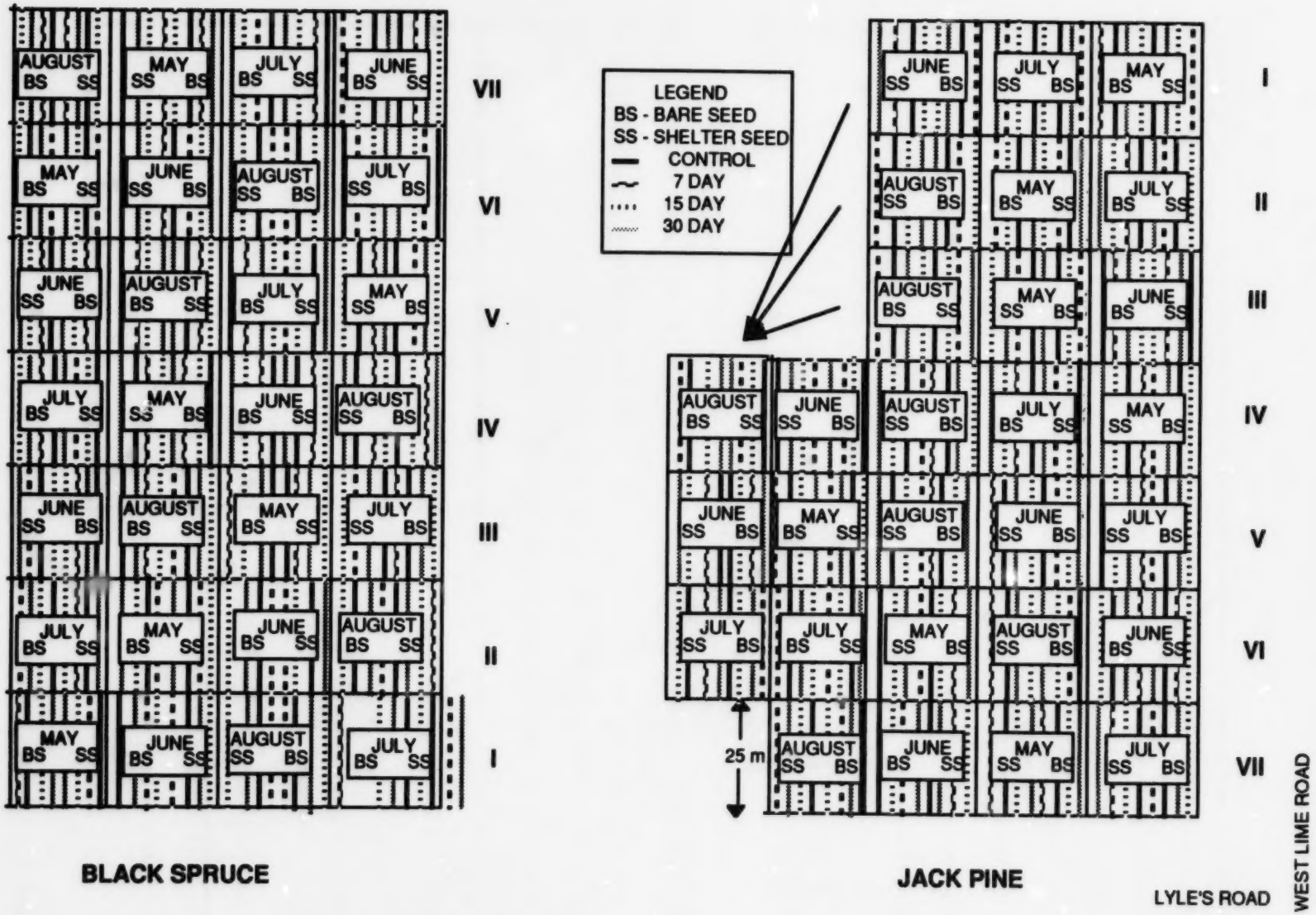

Figure 2. Layout of 1989 stratified seeding trial.

Third year assessments of the number of germinants and height of the tallest seedling per spot were done September 1991 for black spruce only. Second year results for both species were collected in the fall of 1990. Height was measured as the distance between the soil interface and the top of the stem to the nearest half centimetre. Current annual increment was also determined.

Analysis of variance, after arcsin squareroot transformation of percent stocking values, and Student-Newman-Keuls (SNK) tests were used to determine significant differences (Steele and Torrie 1980).

1990 Field Trial

In 1990, the trial was located in the same general area as the 1989 trial (Figure 1) but on a NWO FEC soil type S1 (dry sand). The area, which previously supported a mixedwood, was cut in 1988 and site prepared with a disc trencher in 1989. The area was lightly burned in the fall of 1989 and came back heavily to raspberry, grass and herbaceous ground cover.

Sowing was done as in 1989 but only four blocks were included in the design. The layout of this trial is shown in Figure 3. Seeds were sown on May 16, July 10 and August 14. Conditions were wet and cool in May but warm and sunny for the latter two sowing times.

Second year assessments were done in 1991 using the same parameters as for the 1989 trial. Data analysis paralleled that used for the 1989 trial.

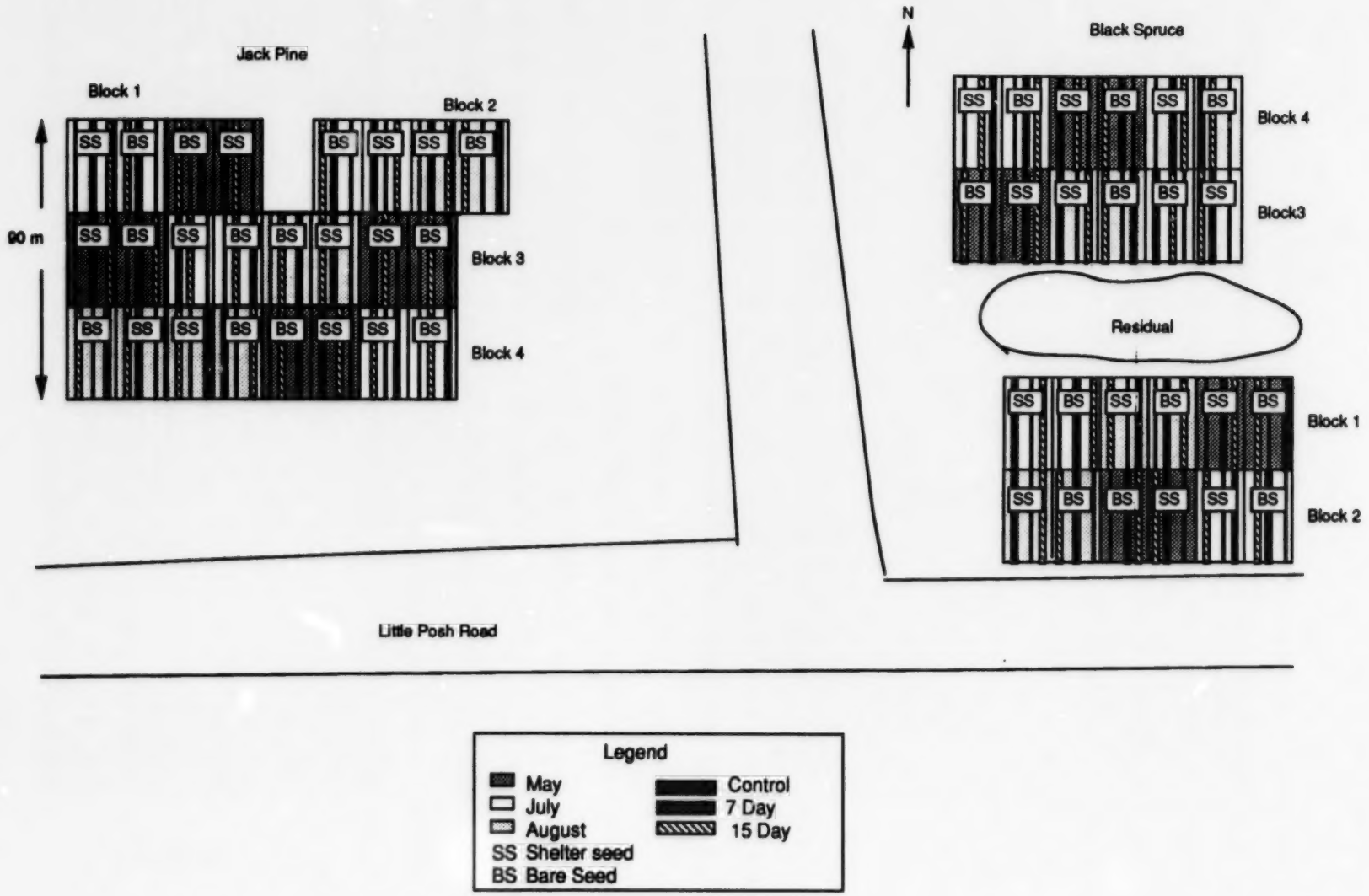

Figure 3. Layout of 1990 stratified seeding trial.

Germination Tests

Germination tests were carried out immediately and three weeks (1990 only) after stratification for 0, 7, 15, and 30 days (1989 only) to determine the effect of storage on the rate and amount of germination. Four replicates of 100 seeds from each stratification treatment for each of black spruce and jack pine were included. Seeds were germinated on Kimpak paper in petri dishes at room temperature and ambient light conditions. Stored seeds were air dried, bagged and kept refrigerated.

Germinated seed was tallied daily. Cumulative percent germination at five day intervals was subjected to analysis of variance after arcsin squareroot transformation of percent values (Steele and Torrie 1980).

RESULTS

Results summarized are third year measurements for black spruce established in 1989 and second year for all others.

1989 Trial

Black Spruce - Third Year Results

Percent stocked spots was significantly higher for the May and June sowings than for the later sowings for bare seeded black spruce (Figure 4). Average stocking levels were very low ranging from 4-30%. There were no significant differences in stocking levels between stratification treatments within sowing times.

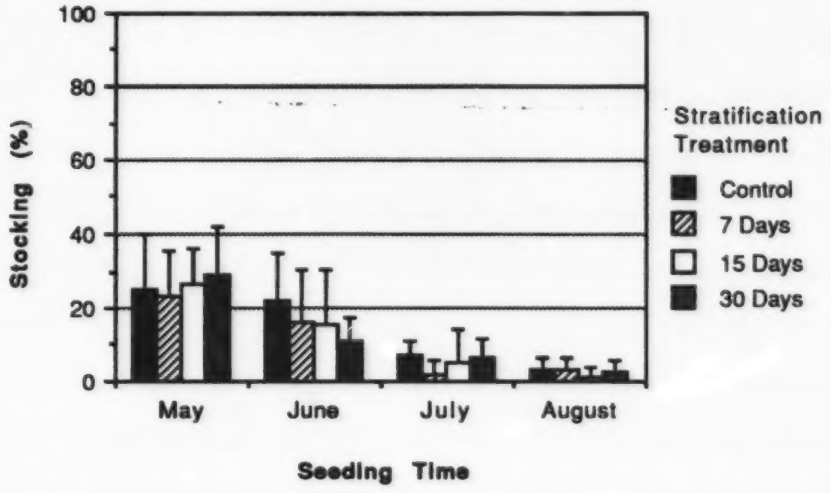

Figure 4. Percent stocked spots for bare seeded black spruce after three growing seasons.

Percent stocked spots were significantly higher for shelter seeded black spruce ranging from 55-85% (Figure 5) but there were no significant differences between stratification treatments. Although the percent stocked spots decreased from May to August sowings, the difference was not significant.

The average number of germinants per spot for bare seeded black spruce was highest in May and lowest in July and August (Figure 6). There were no significant differences in the number of germinants per spot between stratification treatments within sowing times. The average number of germinants per spot for bare seeded black spruce was less than one out of ten seeds sown.

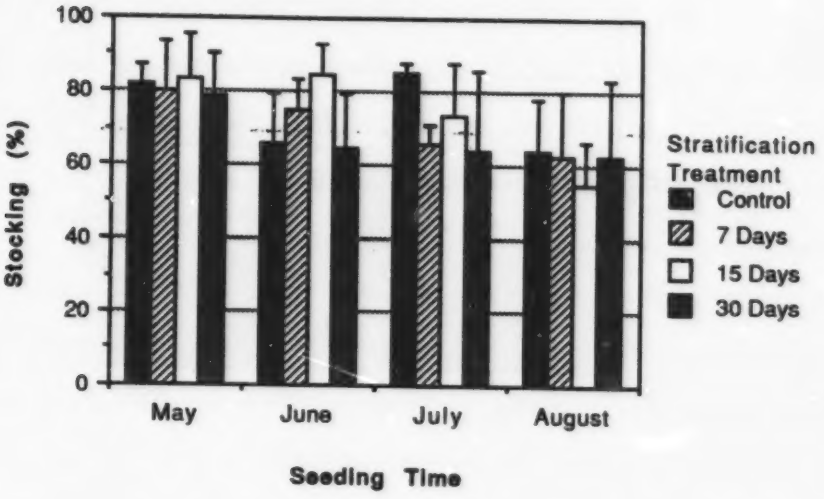

Figure 5. Percent stocked spots for shelter seeded black spruce after three growing seasons.

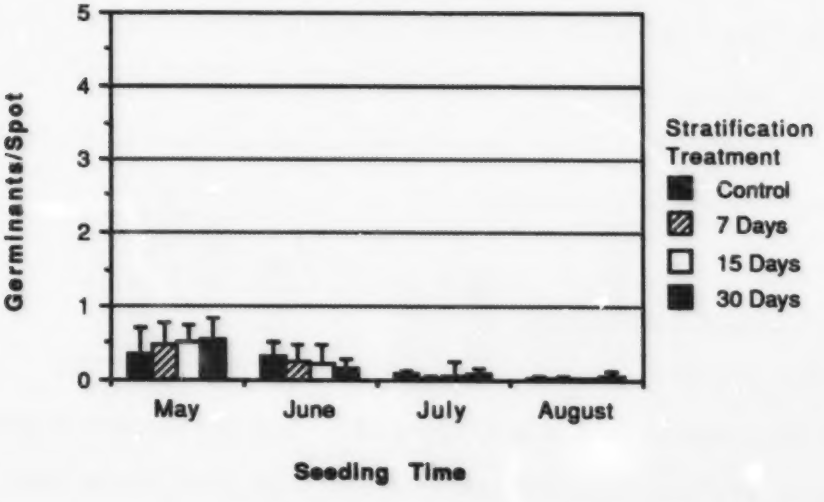

Figure 6. Germinants per spot (of 10 sown) for bare seeded black spruce after three growing seasons.

There were no significant differences in the number of germinants per spot between stratification treatments for shelter seeded black spruce (Figure 7). The August sowing time had significantly fewer germinants per spot. The average number of germinants per spot was two out of five seeds sown. Shelter cones had significantly more germinants per spot than bare seed over all sowing times despite fewer seeds sown.

The average number of germinants per stocked spot for bare seeded black spruce was 1.5 with no difference between stratification treatments (Figure 8). The number decreased across sowing times from 1.8 in May to 1.2 in August. There was an average of three seedlings per stocked spot in the shelter cones with no significant differences between sowing times or stratification treatments (Figure 9).

Average height of bare seeded black spruce decreased across sowing times from 6.5 cm to 3.4 cm (Figure 10). The 30 day stratification treatment produced significantly larger seedlings than the other treatments for the May sowing only.

Average seedling height generally decreased across sowing times for shelter seeded black spruce except that seedlings sown in June were taller than those sown in May (Figure 11). There was no significant difference in seedling height between stratification treatments.

Jack Pine - Second Year Results

Percent stocked spots was higher for May through July than for the August sowing for bare seeded jack pine (Figure 12). Overall percent stocked spots was higher for stratified seed than control seed. The differences were significant in May, June and August. Stocking remained below 60% in all cases.

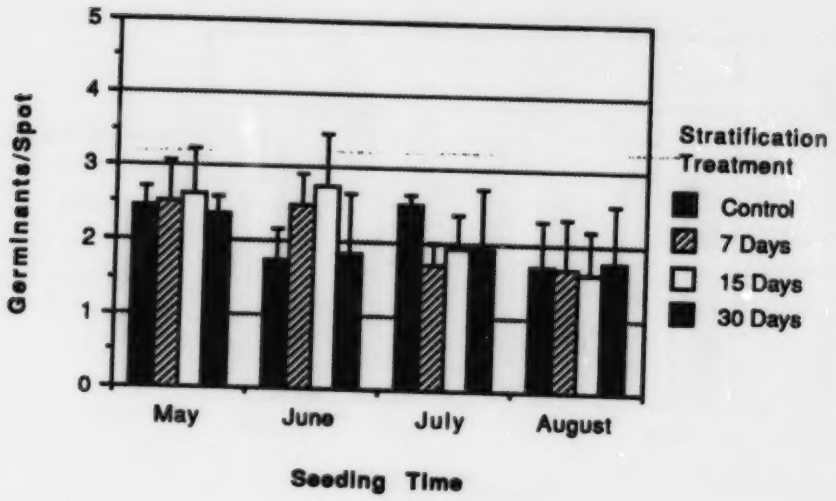

Figure 7. Germinants per spot (of 5 sown) for shelter seeded black spruce after three growing seasons.

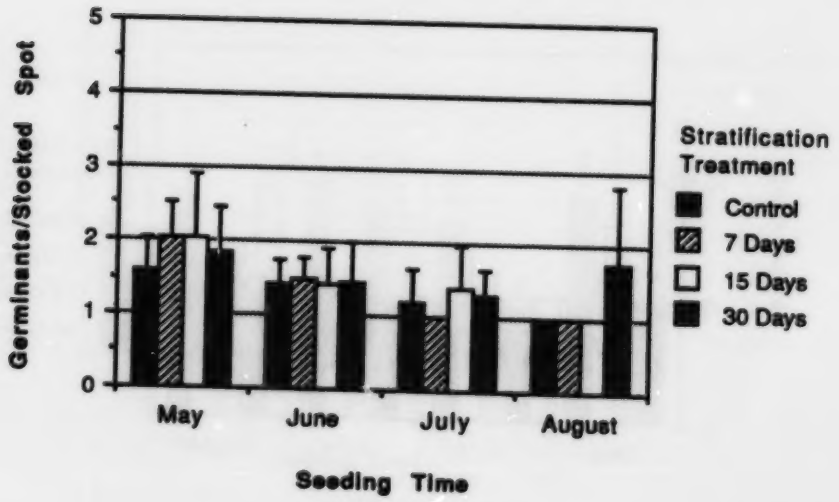

Figure 8. Germinants per stocked spot (of 10 sown) for bare seeded black spruce after three growing seasons.

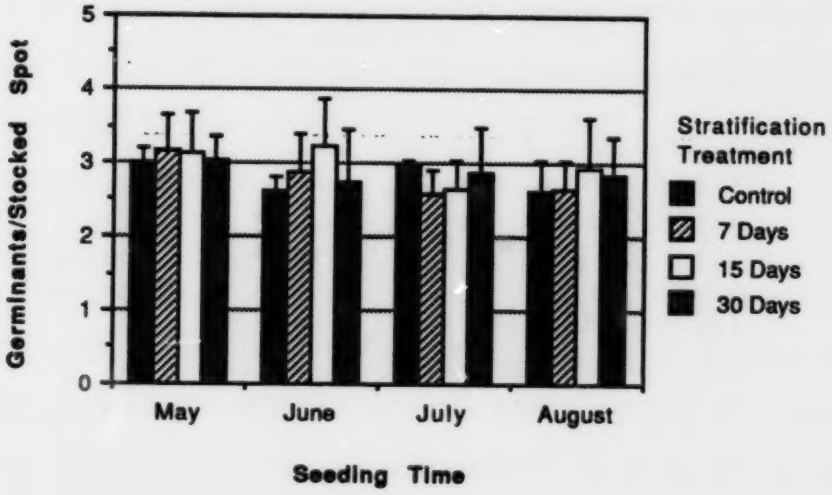

Figure 9. Germinants per stocked spot (of 5 sown) for shelter seeded black spruce after three growing seasons.

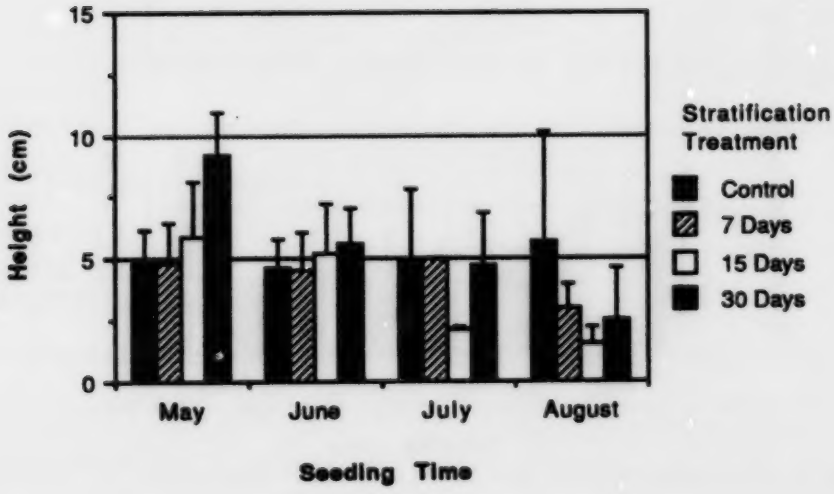

Figure 10. Height of bare seeded black spruce after three growing seasons.

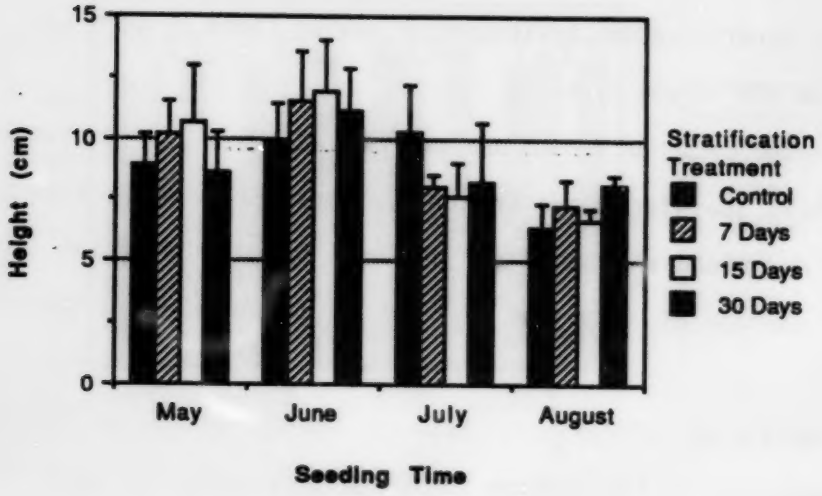

Figure 11. Height of shelter seeded black spruce after three growing seasons.

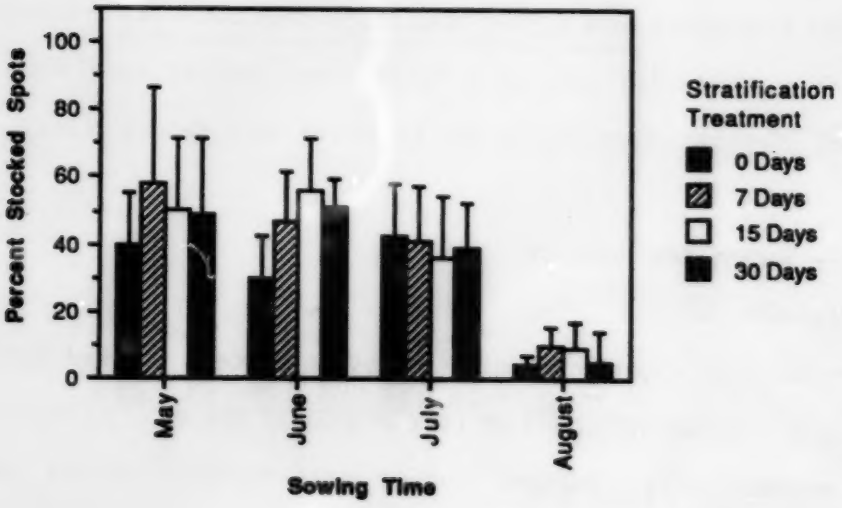

Figure 12. Percent stocked spots for bare seeded jack pine after two growing seasons.

For shelter seeded jack pine there were no differences in percent stocked spots between stratification treatments or sowing times (Figure 13). Stocking remained above 80% in all cases.

The average number of germinants per spot at the end of the second growing season for bare seed for the four sowing times is shown in Figure 14. No significant differences were apparent between stratification treatments. Overall, an average of less than one out of ten bare seeds germinated and survived.

No significant differences between sowing times or stratification treatments occurred in the average number of germinants per spot for shelter seeded jack pine at the end of the second growing season (Figure 15). An average of three to four out of five seeds per shelter cone germinated and survived.

The average height of bare seeded and shelter seeded jack pine decreased across sowing times (Figures 16 and 17 respectively). Overall, seedlings from 15 and 30 day treatments were taller than those from the 7 day treatment and all seedlings from stratified seed were taller than control seedlings. Shelter sceded stock averaged about double the height of bare seeded stock.

1990 Trial - Second Year Results

Black Spruce

There were no significant differences in percent stocked spots between stratification treatments for either bare or shelter seeded black spruce (Figures 18 and 19 respectively). Percent stocked spots decreased across sowing times for bare seeded black spruce but remained stable for shelter seeded spruce. Stocking remained below 20% for bare seeded black spruce and ranged from 50 - 75% for shelter seeded spruce.

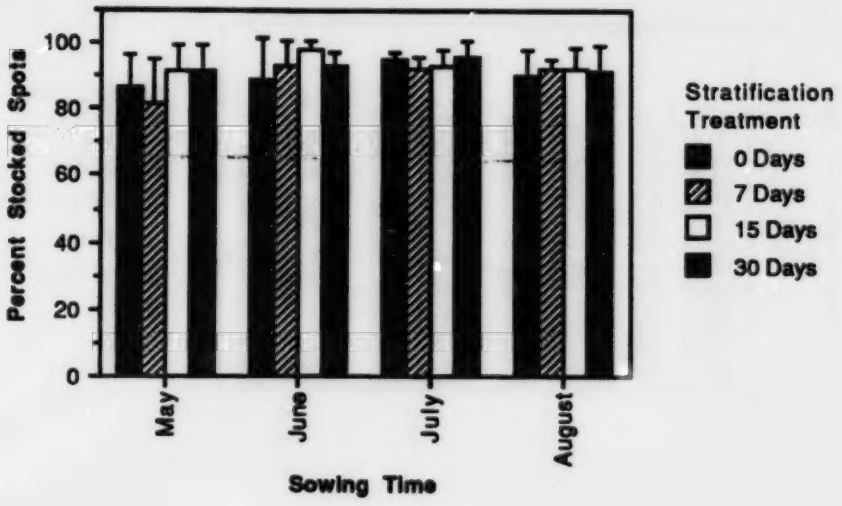

Figure 13. Percent stocked spots for shelter seeded jack pine after two growing seasons.

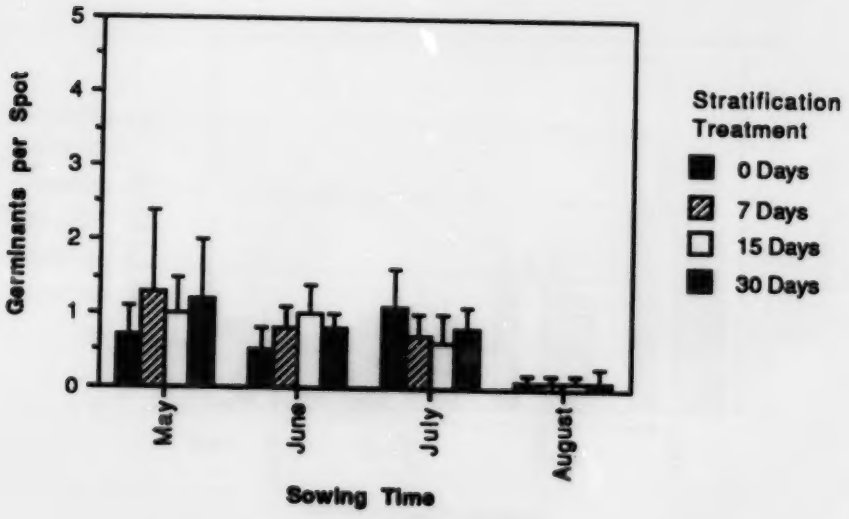

Figure 14. Germinants per spot (of 10 sown) for bare seeded jack pine after two growing seasons.

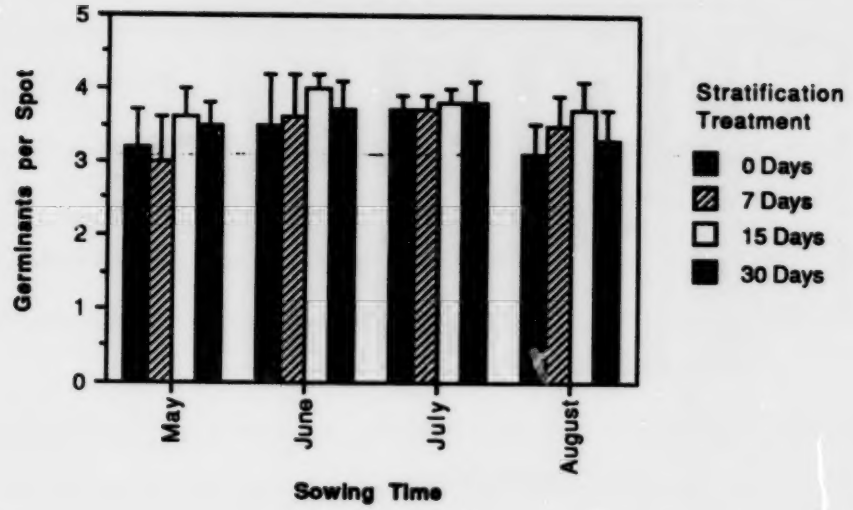

Figure 15. Germinants per spot (of 5 sown) for shelter seeded jack pine after two growing seasons.

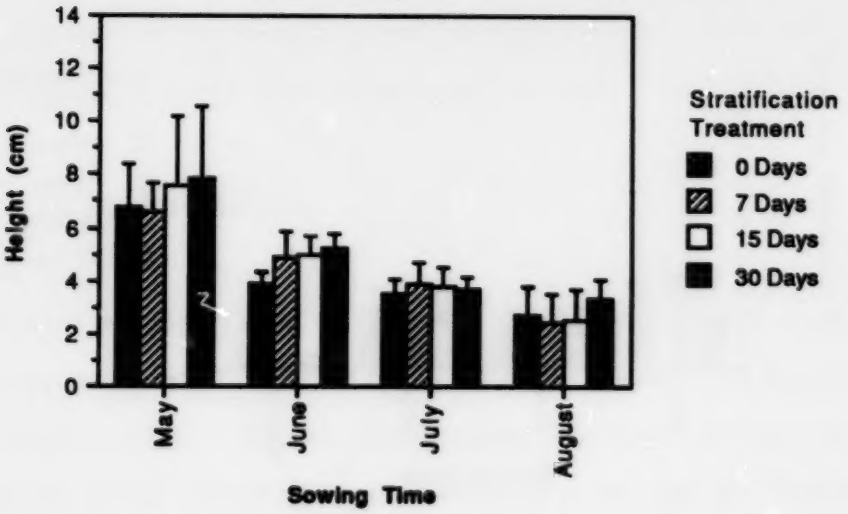

Figure 16. Height of bare seeded jack pine after two growing seasons.

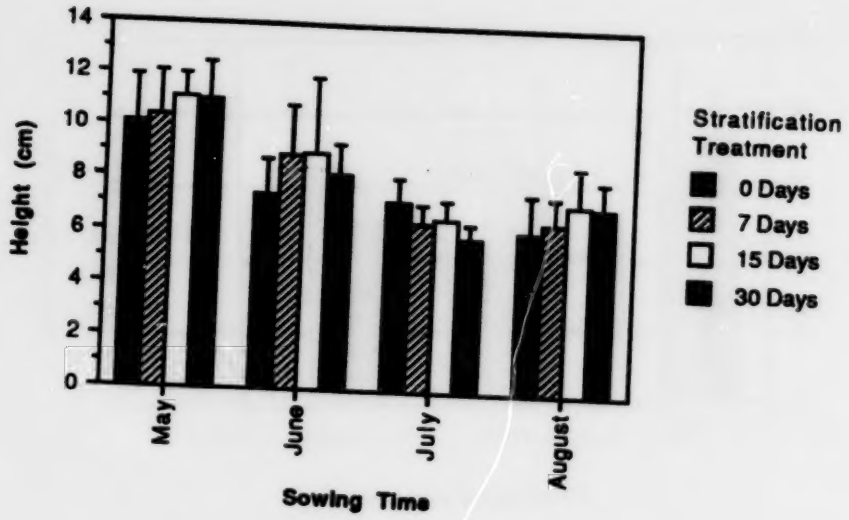

Figure 17. Height of shelter seeded jack pine after two growing seasons.

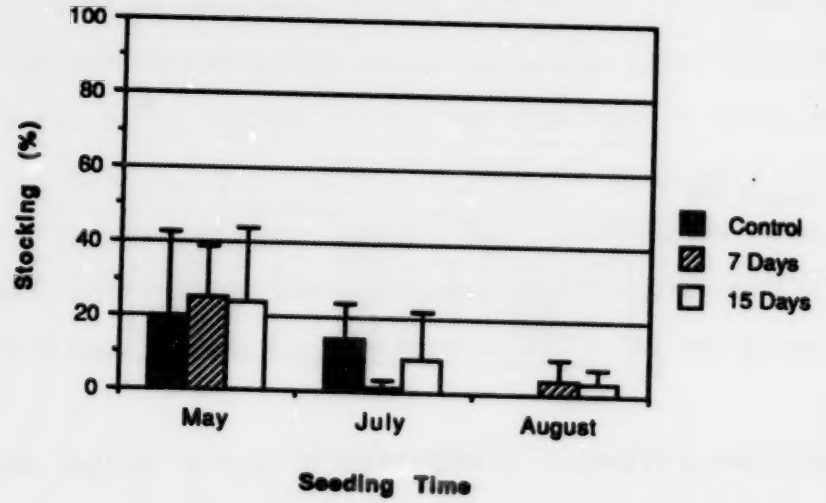

Figure 18. Percent stocked spots for bare seeded black spruce after two growing seasons.

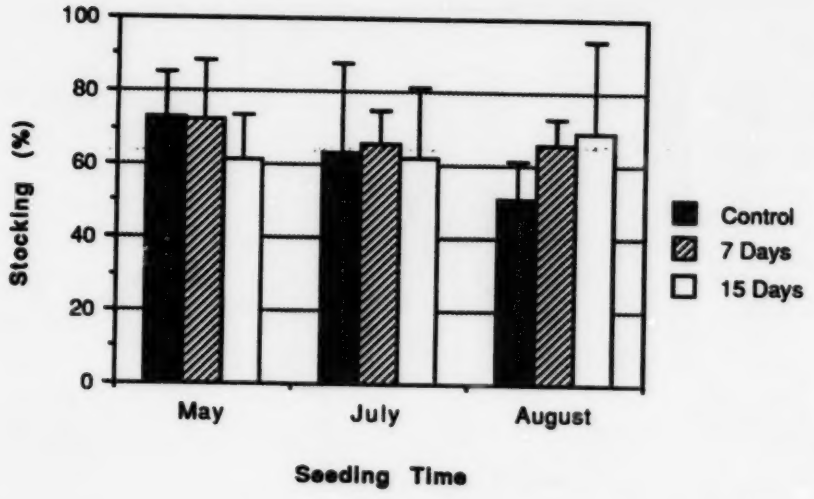

Figure 19. Percent stocked spots for shelter seeded black spruce after two growing seasons.

No differences in the number of germinants per spot was found between stratification treatments regardless of sowing method (Figures 20 and 21). Bare seeded spruce sown in May had the most germinants per spot and August the least. The average was less than 1 seedling per spot. Sowing time did not significantly influence the number of germinants per spot for shelter seeded spruce with the average being 1.5 seedlings per spot.

The number of germinants per stocked spot did not differ between stratification treatments or sowing times for either bare or shelter seeded black spruce (Figures 22 and 23). The averages were 1.5 and 2.0 seedlings per spot respectively.

No significant differences in the height of bare or shelter seeded black spruce were found between stratification treatments (Figures 24 and 25 respectively). Height decreased significantly across sowing times for both bare and shelter seeded stock. Again, shelter seeded stock averaged almost twice the height of bare seeded stock.

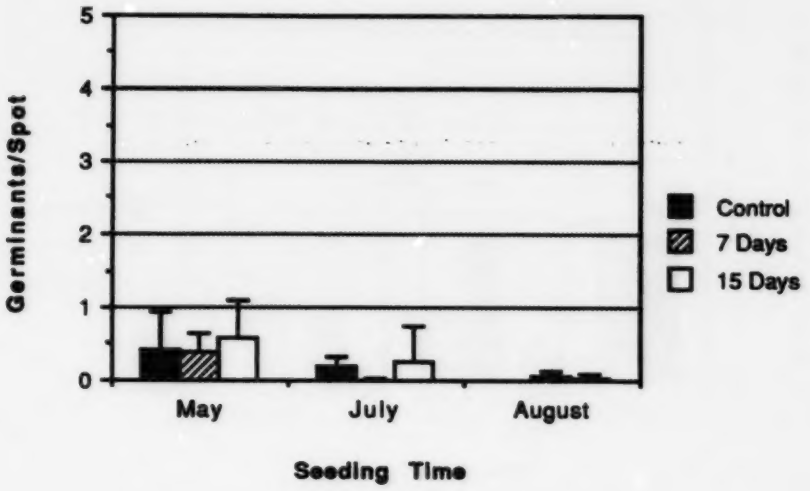

Figure 20. Germinants per spot (of 10 sown) for bare seeded black spruce after two growing seasons.

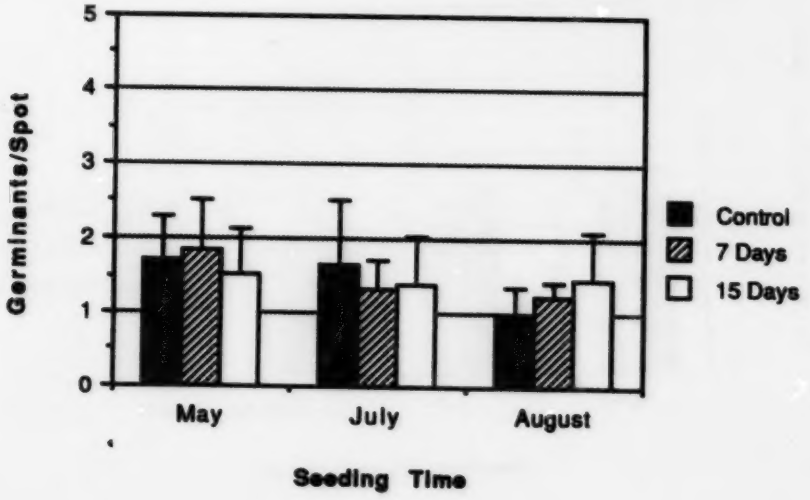

Figure 21. Germinants per spot (of 5 sown) for shelter seeded black spruce after two growing seasons.

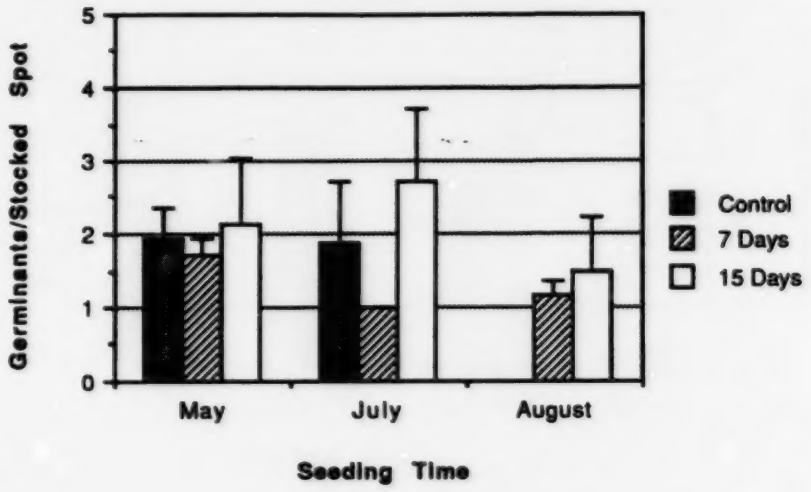

Figure 22. Germinants per stocked spot (of 10 sown) for bare seeded black spruce after two growing seasons.

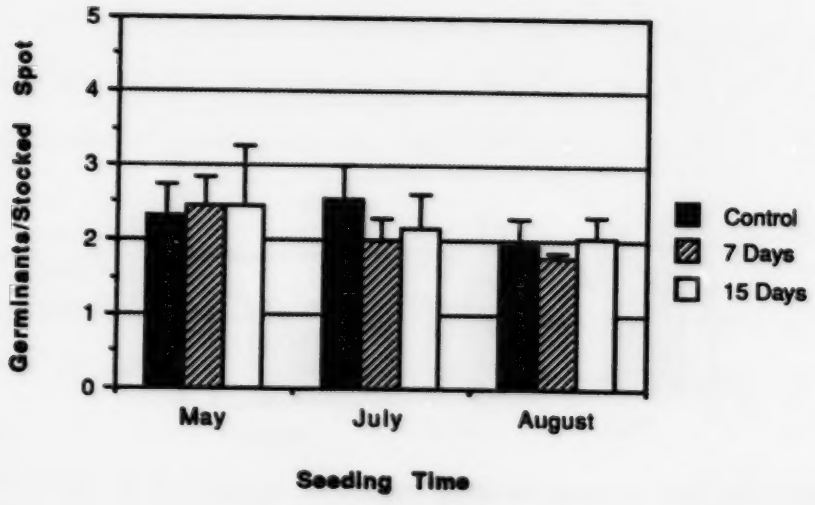

Figure 23. Germinants per stocked spot (of 5 sown) for shelter seeded black spruce after two growing seasons.

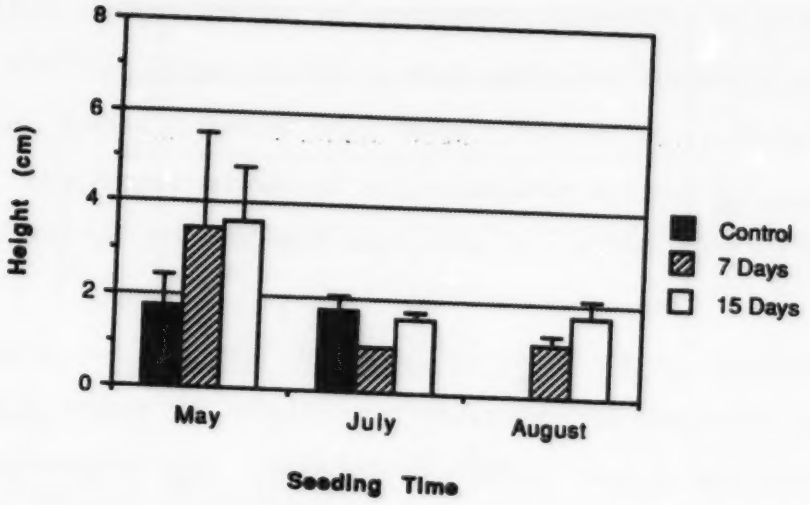

Figure 24. Height of bare seeded black spruce after two growing seasons.

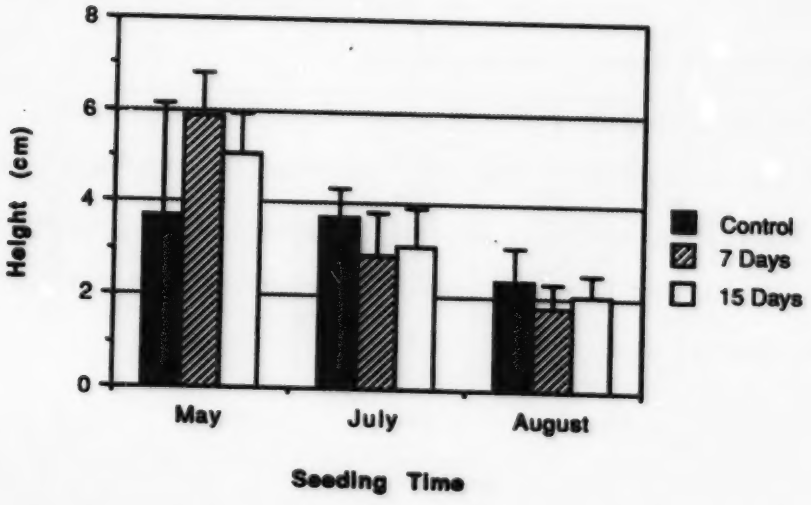

Figure 25. Height of shelter seeded black spruce after two growing seasons.

Jack Pine

There were no significant differences in percent stocked spots between stratification treatments for either bare or shelter seeded jack pine (Figures 26 and 27 respectively). Percent stocked spots decreased across sowing times for bare seeded jack pine but remained stable for shelter seeded pine. Stocking averaged <20% for bare seeded pine and ranged from 50 - 75% for shelter seeded pine.

No differences in the number of germinants per spot was found between stratification treatments for bare seeded pine (Figure 28). Number of germinants decreased across sowing times for bare seeded pine. Fewer germinants per spot were found under shelter cones for control than for stratified seed (Figure 29). Sowing time did not influence the number of germinants per spot for shelter seeded pine.

The number of germinants per stocked spot did not differ significantly between treatments for either bare or shelter seeded pine but did decrease slightly across sowing times (Figures 30 and 31).

The height of jack pine differed significantly between sowing times with May sown seedlings being taller than July sown seedlings for bare seeded stock and July and August sown seedlings for shelter seeded stock (Figures 32 and 33 respectively). Stratified bare seed sown in May resulted in significantly taller seedlings than control seed.

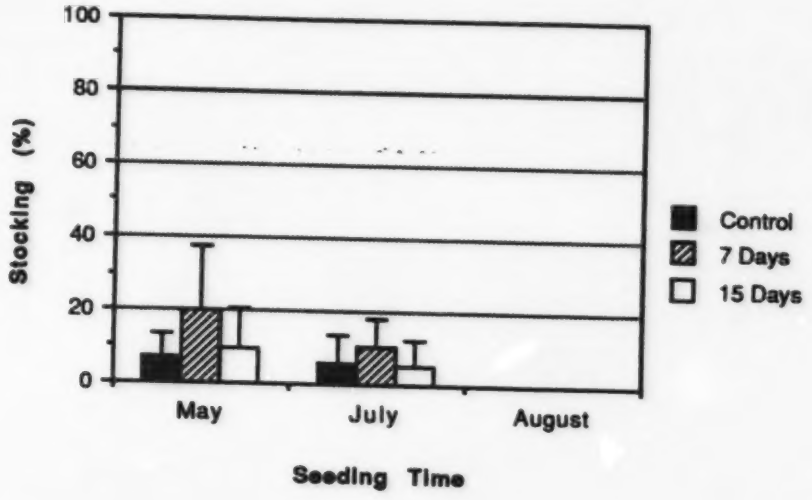

Figure 26. Percent stocked spots for bare seeded jack pine, established in 1990, after two growing seasons.

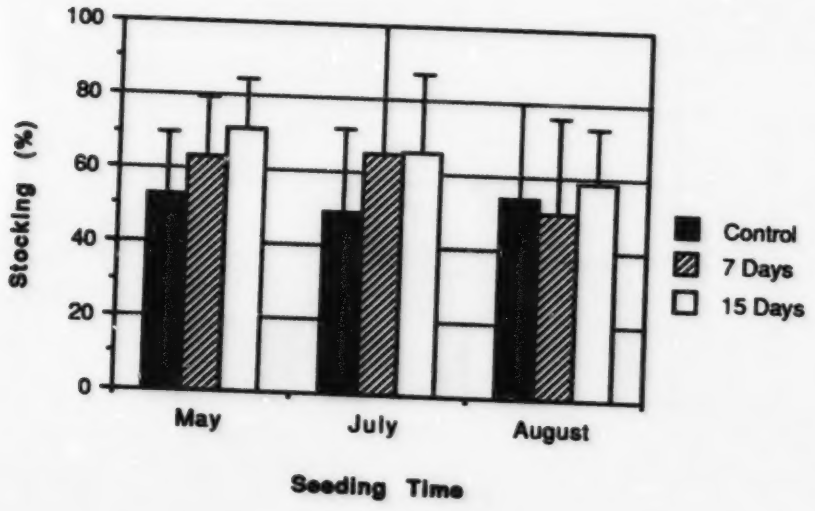

Figure 27. Percent stocked spots for shelter seeded jack, established in 1990, after two growing seasons.

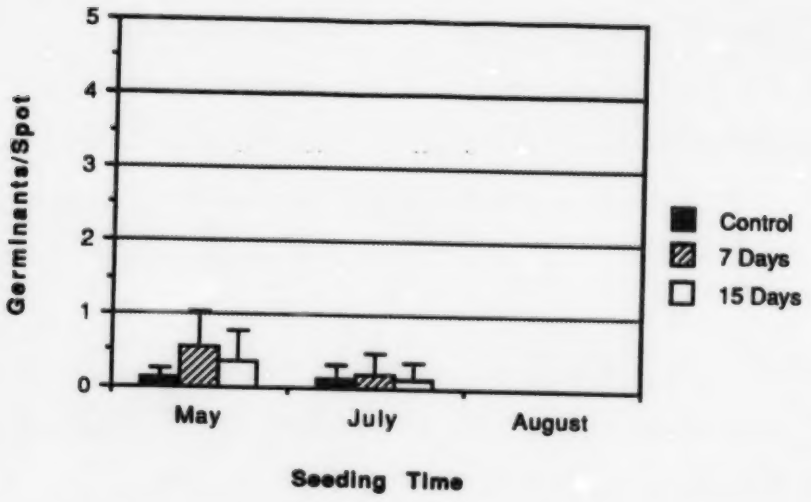

Figure 28. Germinants per spot (of 10 sown) for bare seeded jack pine, established in 1990, after two growing season.

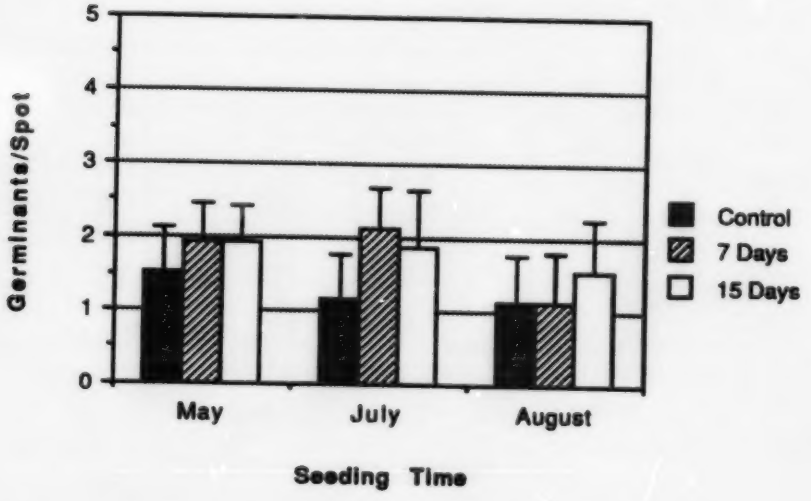

Figure 29. Germinants per spot (of 5 sown) for shelter seeded jack pine, established in 1990, after two growing seasons.

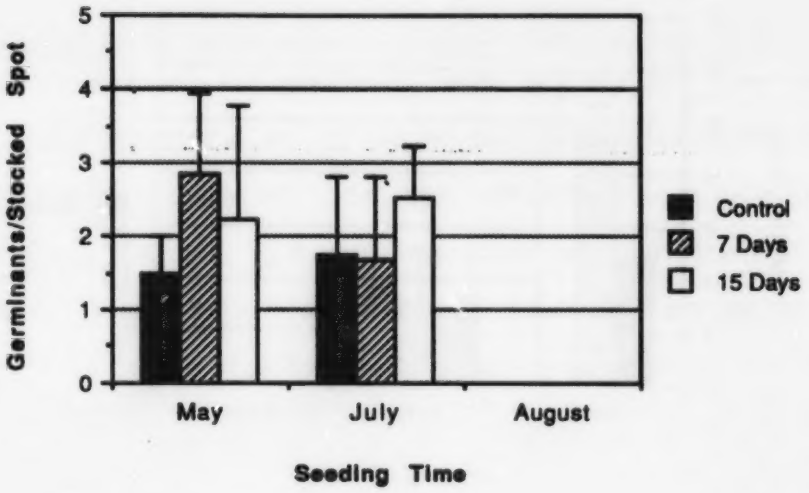

Figure 30. Germinants per stocked spot (of 10 sown) for bare seeded jack pine, established in 1990, after two growing seasons.

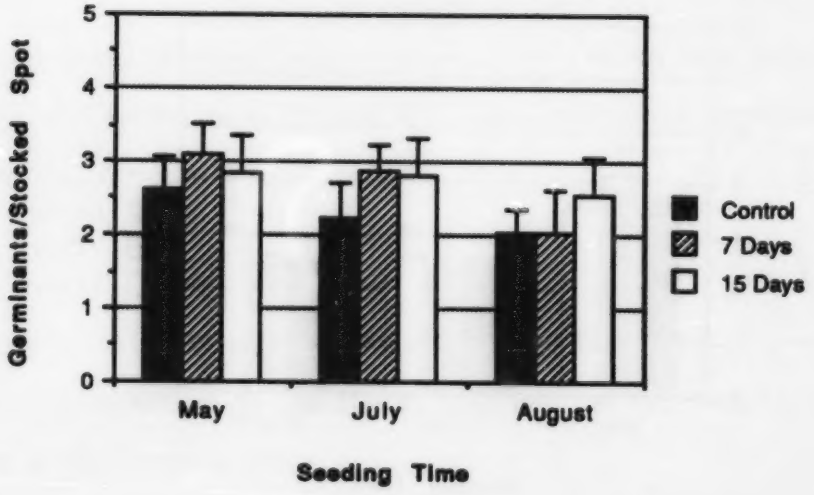

Figure 31. Germinants per stocked spot (of 5 sown) for shelter seeded jack pine, established in 1990, after two growing seasons.

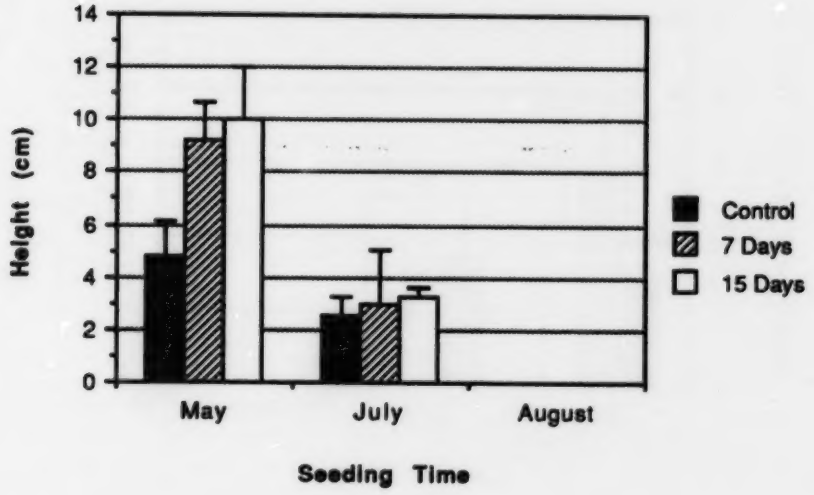

Figure 32. Height of bare seeded jack pine, established in 1990, after two growing seasons.

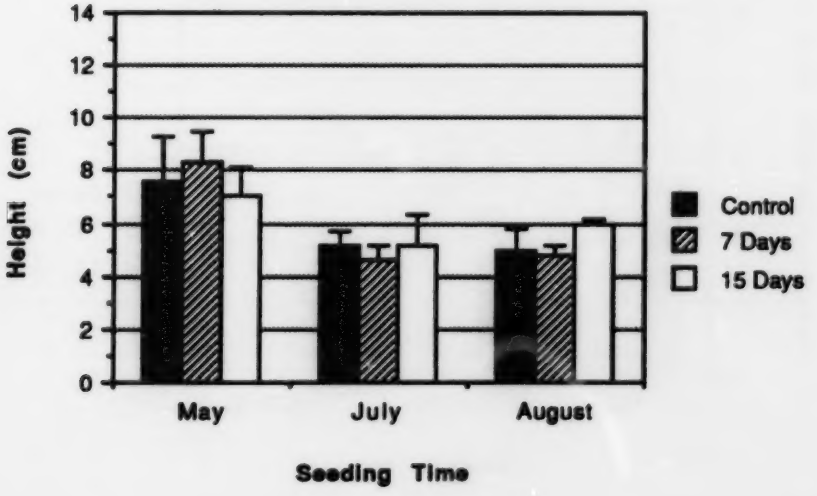

Figure 33. Height of shelter seeded jack pine, established in 1990, after two growing seasons.

Germination Tests

Black Spruce

In 1989, stratified seed germinated more quickly and completely than control seed. Seed stratified for 15 and 30 days germinated faster than that stratified for 7 days (Figure 34). In 1990, stratified seed germinated more quickly than control seed but, by day 10, there were no significant differences between treatments (Figure 35). Storage did not reduce the germinative capacity of the seed although it did slow down germination by a few days.

Jack Pine

In 1989, stratified seed germinated more quickly and completely than control seed with no significant differences occurring between stratification treatments. The majority of seed germinated between 10 and 15 days from the start of the test. After 35 days only 70% of the control seed had germinated but 95% of the stratified seed had germinated (Figure 36). In 1990, stratified seed was not affected by three weeks of storage but control seed germinated more slowly during the second test (Figure 37). All treatments germinated quickly and close to completely within 10 days.

DISCUSSION

Although stratification treatments increased the speed of germination in both germination tests and in the field, they had limited long-term impact on survival and growth in this study.

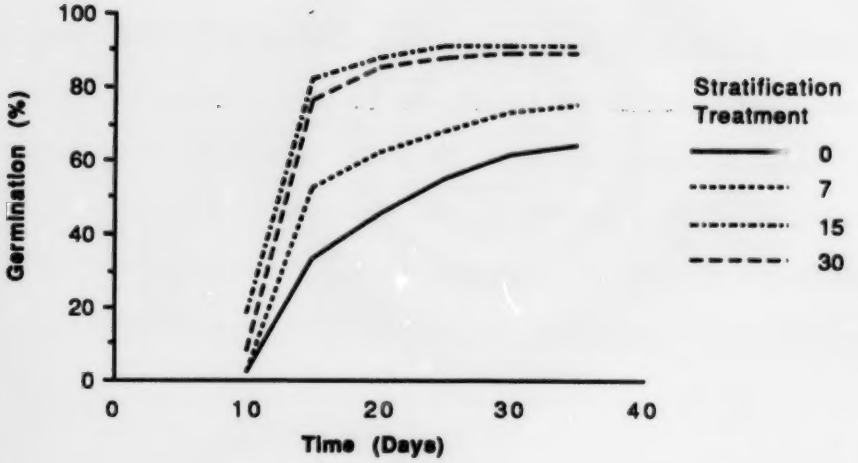

Figure 34. Germination of stratified versus control black spruce seed in 1989 test.

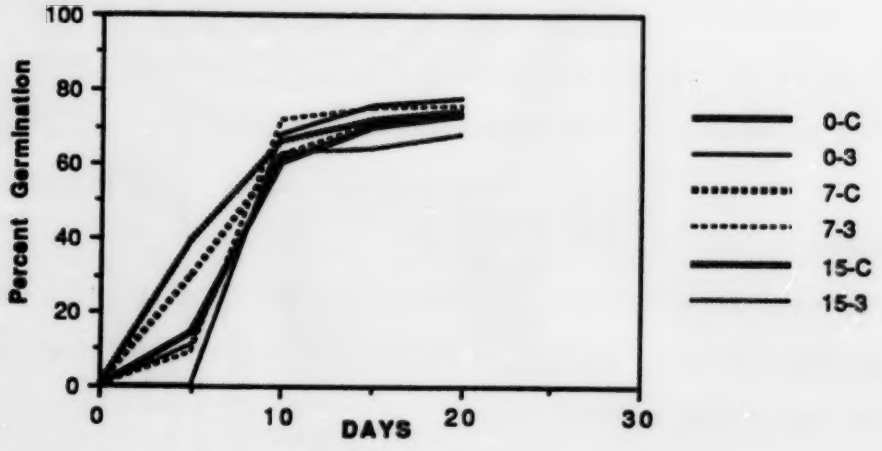

Figure 35. Germination of black spruce seed immediately (C) and three weeks (3) after stratification in 1990 test.

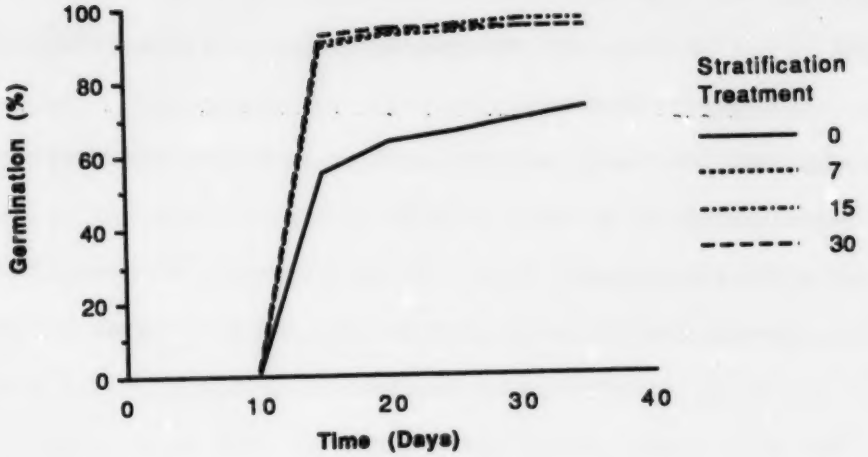

Figure 36. Germination of stratified versus control jack pine seed in 1989 test.

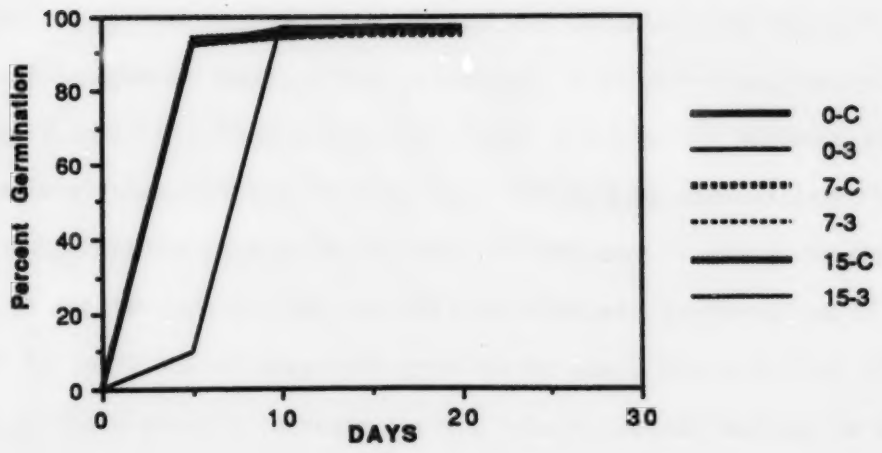

Figure 37. Germination of jack pine seed immediately (C) and three weeks (3) after stratification in 1990 test.

Lahde (1974) found that germination of scots pine (Pinus sylvestris) in field trials was not improved by stratification for 14 days at 4°C. In this study, stocking was increased by stratification for jack pine bare seed sown in May, June, and August in 1989 only.

The number of germinants per spot was increased by stratification for jack pine sown under shelter cones only in 1990.

Myland and Horvath (1988) found that height growth response at the end of the first season depended on seed source and not stratification treatment. In this study, the 30 day stratification treatment resulted in larger black spruce seedlings for bare seeded stock sown in May of 1989 only. For jack pine, stratification increased the size of shelter seedlings in the 1989 trial and bare seedlings sown in May of 1990 only. Boyer et al. (1985) found that stratification increased the diameter of loblolly pine at the end of the first season in the nursery. Unfortunately, diameter was not assessed in this study.

Shelter cones reduced the number of seeds required and resulted in better germination and establishment for both species. Seed is less subject to washing, predation and smothering under the cones. Lahde (1974) found that shelter cones improved germination by 15 - 33%. Dominy and Wood (1986) and Lahde (1974) attributed better stocking of black spruce and jack pine under shelter cones to higher humidity and air temperatures, less frost heaving and protection from seed predation and browsing. In both the 1989 and 1990 trials, percent stocked spots for both jack pine and black spruce were increased in the order of 50% through the use of shelter cones.

Dominy and Wood (1986) did not find that shelter cones significantly improved the height growth of black spruce or jack pine after five years except for one of their June sowings. In this study, height of shelter seedlings was generally double that of bare seedlings except for stratified jack pine seed sown in May in the 1990 trial.

May and June sown seedlings performed best with success decreasing as the season progressed in both years. Other trials have also found spring seeding to give the best results. Stratified seed did poorly in July. It may be that stratification allows seed to germinate when environmental conditions (temperature and moisture) are less than optimal and because germination is more uniform, all of the seedlings end up dying if poor conditions continue. Control seeds do not germinate uniformly and therefore, if some die initially, others remain that can establish during the next favourable period (Downie, pers. comm. 1992).

CONCLUSIONS AND RECOMMENDATIONS

Seed stratification increased the speed of germination in the field but did not impact on overall seeding success except in a few situations. Stratification may increase percent stocked spots for bare seeded jack pine under certain conditions and may increase the initial growth of seedlings. The inconsistent results between years in this trial indicates that stratification cannot be relied upon to improve seeding success.

Stratification cannot be used to extend the seeding window; in fact, control seed performed better in July than stratified seed. A mixture of stratified and unstratified seed may provide the best results in uncertain environmental conditions. Direct seeding should be done in the spring if at all possible.

Shelter cones greatly increase seeding success for both black spruce and jack pine. Four seeds per cone are probably adequate for spruce and three for jack pine, although this will still result in more than one seedling per stocked spot with some spots remaining unstocked. Seeding success can be expected to vary by year and decreasing the number of seeds too much will decrease stocking.

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